IN THE SPECIFICATION

Please amend pages 1, 2, 3, 4, 8 and 9 as follows:

[redacted]

SERINE PROTEASE INHIBITOR

The invention relates to a serine protease inhibitor comprising an ether bonded arginine replacement, a pharmaceutical composition containing the same, as well as the use of said serine protease inhibitor for the manufacture of a medicament.

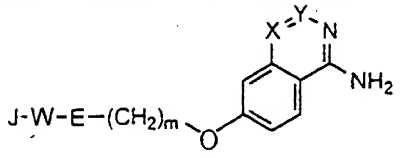
Serine proteases are enzymes which play an important role in the blood coagulation cascade. Apart from thrombin and factor Xa, other examples of this group of proteases comprise the factors VIIa, IXa, XIa, XIIa, and protein C.

Thrombin is the final serine protease enzyme in the coagulation cascade. The prime function of thrombin is the cleavage of fibrinogen to generate fibrin monomers, which are cross-linked to form an insoluble gel. In addition, thrombin regulates its own production by activation of factors V and VIII earlier in the cascade. It also has important actions at cellular level, where it acts on specific receptors to cause platelet aggregation, endothelial cell activation and fibroblast proliferation. Thus thrombin has a central regulatory role in haemostasis and thrombus formation. Since inhibitors of thrombin may have a wide range of therapeutic applications, there is a continuous effort to find new serine protease inhibitors. The difficulty in this endeavour is to find a compound in which the properties of therapeutic safety, selectivity, potency and synthetic accessibility are combined. In particular the bioavailability with the oral route of administration can be problematic for selective thrombin inhibitors.

In WO 97/16444 thrombine inhibitors are described with modifications of the tripeptide sequence phenylalanyl-prolyl-arginine, in which a carboxyl group is present at the amino terminal and arginine is replaced by a basic (aminoiminomethyl)phenyl or a basic (aminoiminomethyl)pyridinyl group linked to a prolyl analogue by an ether bond.

It is the object of this invention to provide new chemically accessible serine protease inhibitors with a favourable pharmacological and toxicological profile with sufficient potency for therapeutic use.

It has been found that this object can be met with a serine protease inhibitor, and in particular a thrombin inhibitor, having the formula (I),



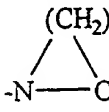
Formula (I)

in which

J is H, R^1, R^1-O-C(O)-, R^1-C(O)-, R^1-SO_2-, R^3OOC-$(CHR^2)_p$-, $(R^{2a},R^{2b})N$-CO-$(CHR^2)_p$- or Het-CO-$(CHR^2)_p$-;

W is an amino-acid of the formula -NH-CHR^1-C(O)-, -NR^4-CH[$(CH_2)_qC(O)OR^1$]-C(O)-, -NR^4-CH[$(CH_2)_qC(O)N(R^{2a},R^{2b})$]-C(O)-, -$NR^4$-CH[$(CH_2)_qC(O)$Het]-C(O)-, D-1-Tiq, D-3-Tiq, D-Atc, Aic, D-1-Piq or D-3-Piq;

E is -NR^2-CH_2- or the fragment



, optionally substituted with (1-6C)alkyl, (1-6C)alkoxy or benzyloxy;

R^1 is selected from (1-12C)alkyl, (2-12C)alkenyl, (2-12C)alkynyl, (3-12C)cycloalkyl and (3-12C)cycloalkyl(1-6C)alkylene, which groups may optionally be substituted with (3-12C)cycloalkyl, (1-6C)alkoxy, oxo, OH, CF_3 or halogen, and from (6-14C)aryl, (7-15C)aralkyl, (8-16C)aralkenyl and (14-20C)(bisaryl)alkyl, whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-12C)cycloalkyl, (1-6C)alkoxy, OH, CF_3 or halogen;

R^2, R^{2a} and R^{2b} are each independently selected from H, (1-8C)alkyl, (2-8C)alkenyl, (2-8C)alkynyl, (3-8C)cycloalkyl and (3-6C)cycloalkyl(1-4C)alkylene, which can each be optionally substituted with (3-6C)cycloalkyl, (1-6C)alkoxy, CF_3 or halogen, and from (6-14C)aryl and (7-15C)aralkyl whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-6C)cycloalkyl, (1-6C)alkoxy, CF_3 or halogen;

R^3 is as defined for R^2 or Het-(1-6C)alkyl;

R^4 is H or (1-3C)alkyl;

X and Y are CH or N with the proviso that they are not both N;

Het is a 4-, 5- or 6-membered heterocycle containing one or more heteroatoms selected from O, N and S;

m is 1 or 2;

p is 1, 2 or 3;

q is 1, 2 or 3;

t is 2, 3 or 4;

and prodrugs thereof;

and pharmaceutically acceptable addition salts and/or solvates thereof.

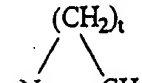
In particular, the compounds of the invention may possess improved bioavailability after oral administration.

In preferred embodiments of this invention compounds have formula (I) in which

J is H, R^1, R^1-SO_2-, R^3OOC-$(CHR^2)_p$-, $(R^{2a},R^{2b})N$-CO-$(CHR^2)_p$- or Het-CO-$(CHR^2)_p$-; more preferred is (3-12C)cycloalkyl optionally substituted with (1-6C)alkoxy; or

W is an amino-acid of the formula -NH-CHR^1-C(O)-, -NR^4-CH[(CH$_2$)$_q$C(O)OR^1]-C(O)-, -NR^4-CH[(CH$_2$)$_q$C(O)N(R^{2a},R^{2b})]-C(O)-, -NR^4-CH[(CH$_2$)$_q$C(O)Het]-C(O)-; more preferred are an amino acid of the formula -NH-CHR^1-C(O)- or an L-amino acid of the formula -NR^4-CH[(CH$_2$)$_2$C(O)N(R^{2a},R^{2b})]-C(O)-; or

E is -N(3-6C)cycloalkyl-CH$_2$- or the fragment



, optionally substituted with (1-6C)alkyl, (1-6C)alkoxy or benzyloxy; more preferred is without substitution; or

R^1 is selected from (1-12C)alkyl, (3-12C)cycloalkyl and (3-12C)cycloalkyl(1-6C)alkylene, which groups may optionally be substituted with (3-12C)cycloalkyl, (1-6C)alkoxy, or oxo and from (6-14C)aryl, (7-15C)aralkyl and (14-20C)(bisaryl)alkyl, whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-12C)cycloalkyl, (1-6C)alkoxy, OH, CF_3 or halogen; more preferred is a selection from (3-12C)cycloalkyl and (3-12C)cycloalkyl(1-6C)alkylene, which groups may optionally be substituted with (3-12C)cycloalkyl or (1-6C)alkoxy, and from (6-14C)aryl, (7-15C)aralkyl and (14-20C)(bisaryl)alkyl, whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-12C)cycloalkyl, (1-6C)alkoxy or halogen; or

R^2 is H; or

R^{2a} and R^{2b} are each independently selected from H, (1-8C)alkyl, (3-8C)cycloalkyl and (3-6C)cycloalkyl(1-4C)alkylene, which can each be optionally substituted with (3-6C)cycloalkyl or (1-6C)alkoxy, and from (6-14C)aryl and (7-15C)aralkyl whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-6C)cycloalkyl, (1-6C)alkoxy, CF_3 or halogen; or

R^3 is selected from H, (1-8C)alkyl, (3-8C)cycloalkyl and (3-6C)cycloalkyl(1-4C)alkylene, which can each be optionally substituted with (3-6C)cycloalkyl or (1-6C)alkoxy, and from (7-15C)aralkyl whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-6C)cycloalkyl, (1-6C)alkoxy, CF_3 or halogen and from Het-(1-6C)alkyl; more preferred is a selection from (1-8C)alkyl and (3-8C)cycloalkyl, which can each be optionally substituted with (3-6C)cycloalkyl or (1-6C)alkoxy, and from (7-15C)aralkyl whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-6C)cycloalkyl, (1-6C)alkoxy, CF_3 or halogen and from Het-(1-6C)alkyl; or

R^4 is H or (1-3C)alkyl; or

X and Y are CH; or

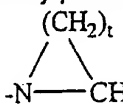
Het is a 4-, 5- or 6-membered heterocycle containing one or more heteroatoms selected from O, N and S; or

m is 2; or

p is 1; or
q is 2; or
t is 3 or 4; more preferred is 4.

In further preferred embodiments W is an amino-acid of the formula -NH-CHR^1-C(O)- or glutamyl [or an (1-6C)alkylester thereof];
R^1 is selected from (3-12C)cycloalkyl and (3-12C)cycloalkyl(1-6C)alkylene, which groups may optionally be substituted with (3-12C)cycloalkyl or (1-6C)alkoxy, and from (6-14C)aryl, (7-15C)aralkyl and (14-20C)(bisaryl)alkyl, whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-12C)cycloalkyl, (1-6C)alkoxy or halogen; and R^3 is selected from (1-8C)alkyl and (3-8C)cycloalkyl, which can each be optionally substituted with (3-6C)cycloalkyl or (1-6C)alkoxy, and from (7-15C)aralkyl whereby the aryl groups may optionally be substituted with (1-6C)alkyl, (3-6C)cycloalkyl, (1-6C)alkoxy, CF_3 or halogen and from Het-(1-6C)alkyl.

Particularly preferred are compounds wherein J is -CH_2COO(1-6C)alkyl, (3-8C)cycloalkyl, -SO_2-10-camphor, -CH_2CONHphenyl or -CH_2CONH(3-8C)cycloalkyl. Other highly preferred compounds are those wherein W is D-cyclohexylalaninyl, D-phenylalaninyl, D-diphenylalaninyl or glutamyl [or an (1-6C)alkylester thereof]. Also particularly preferred are compounds wherein **E** is the fragment



, wherein t is 3 or 4.

Herein the terms have the following meaning:
(1-12C)alkyl, (1-8C)alkyl, (1-6C)alkyl, (1-3C)alkyl means a branched or unbranched alkyl group having 1-12, 1-8, 1-6 and 1-3 carbon atoms, respectively, for example methyl, ethyl, propyl, isopropyl, butyl, sec-butyl, tert-butyl, hexyl, octyl and the like.
(2-12C)alkenyl and (2-8C)alkenyl means a branched or unbranched alkenyl group having 2-12, and 2-8 carbon atoms, respectively, such as ethenyl, 2-butenyl, etc..
(2-12C)alkynyl and (2-8C)alkynyl means a branched or unbranched alkynyl group having 2-12 and 2-8 carbon atoms, respectively, such as ethynyl, propynyl, etc..
(1-6C)alkoxy means an alkoxy group having 1-6 carbon atoms, the alkyl moiety having the meaning as previously defined.
(3-12C)cycloalkyl, (3-8C)cycloalkyl, (3-6C)cycloalkyl means a mono- or bicycloalkyl group having 3-12, 3-8 and 3-6 carbon atoms, respectively, like cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, cyclo-octyl, etc..

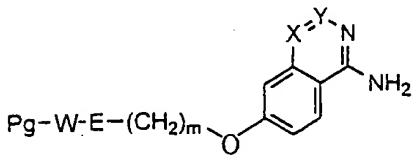
production and handling. Binders are for example cellulose, starches, polyvinylpyrrolidone, and the like.

A suitable lubricant with which the active agent of the invention can be administered is, for example, magnesium stearate.

Surfactants are agents facilitating the contact and migration of compounds in different physical environments such as hydrophilic and hydrophobic environments. Many surfactants are known in the art of making pharmaceutical compositions as for example described in chapter 19 of Remington's Pharmaceutical Sciences (18th edition Editor A.R. Gennaro; Mack Publishing Comp; Easton, Pennsylvania). Surfactants that can be used during the process of preparing the pharmaceutical formulation are, for example, polyethylene glycol (PEG), and the like.

The compounds may also be used with implantable pharmaceutical devices such as those described in US Patent 4,767,628, the contents of which are incorporated by this reference. Then the device will contain sufficient amounts of compound to slowly release the compound (e.g. for more than a month).

Compounds of formula (I) may be prepared from a compound of formula (II), or derivatives thereof wherein the amino group at the aromatic group (arylamino) is protected as urethane such as Alloc or amide such as benzoyl, wherein W, E, X, Y and m have the previously defined meaning and Pg is an N-protecting group (preferably urethane such as Boc).

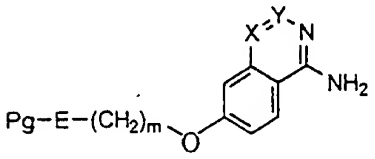


(II)

The term N-protecting group as used in this document means a group commonly used in peptide chemistry for the protection of an α-amino group, like the allyloxycarbonyl (Alloc) group, the *tert*-butyloxycarbonyl (Boc) group, the benzyloxycarbonyl (Z) group, the 9-fluorenylmethyloxycarbonyl (Fmoc) group or the phthaloyl (Phth) group. Removal of the protecting groups can take place in different ways, depending on the nature of those protecting groups. An overview of amino protecting groups and methods for their removal is given in the above mentioned The Peptides, Analysis, Synthesis, Biology, Vol 3. and in T.W. Greene and P.G.M. Wuts, Protective Groups in Organic Synthesis, 2nd edition, 1991, John Wiley & Sons, Inc. .

Removal of the N-protecting group Pg and optional modification(s) of the deprotected amine group using methods known in the field such as peptide coupling, alkylation or reductive amination give compounds of formula (I).

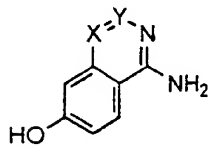
Compounds of formula (II) can be prepared from a compound of formula (III), or derivatives thereof wherein the arylamino is protected as urethane such as Alloc or amide such as benzoyl, wherein E, X, Y, m and Pg have the previously defined meanings. Removal of the N-protecting group Pg in compounds of formula (III) and peptide coupling with compounds of formula Pg-W-OH, in which W and Pg have the previously defined meaning, yields compounds of formula (II).



(III)

Alternatively, compounds of formula (I) can be prepared directly from compounds of formula (III). Removal of the N-protecting group Pg in compounds of formula (III) and a peptide coupling with a compounds of formula J-W-OH, in which J and W have the previously defined meanings and may optionally contain a additional protecting group, afford compounds of formula (I).

Compounds of formulas (I), (II) and (III) are accessible from those of formula (IV) wherein X and Y have the previously defined meanings, or derivatives thereof wherein the arylamino is protected as urethane such as Alloc or amide such as benzoyl, by reaction with alcohols of formula J-W-E-$(CH_2)_m$-OH, Pg-W-E-$(CH_2)_m$-OH or Pg-E-$(CH_2)_m$-OH, in which W, E, J, m and Pg have the previously defined meanings and optionally containing a protecting group, under standard Mitsunobu conditions (tributhylphosphine, dialkyl azodicarboxylate) (R.L. Elliot, H. Kopecka, D.E. Gunn, H.-N. Lin and D.S. Garvey, *Bioorg. Med. Chem. Lett.*, 6, 2283 (1996); K. Wisniewski, A.S. Koldziejczyk and B. Falkiewicz, *J. Pept. Sc.*, 4, 1 (1998)).



(IV)

Alcohols of type formula Pg-E-$(CH_2)_m$-OH in which E = -R^3NCH_2- and m = 2 are accessible by conjugate addition of the corresponding amine R^3NH_2 to ethyl acrylate, reduction of the ester function with lithium aluminium hydride, and subsequent introduction of the N-protecting group Pg.